The spread of cancer is the major reason for cancer-caused deaths. If cancer spread did not occur or if it could be controlled, dramatic reductions would occur in the cancer mortality rate. Localized, nonspreading tumors are usually easily removed or destroyed, in effect curing the individual.

The routes by which cancer spreads are known. Some of the cellular mechanisms that cause cancer spread are also known. Recently, experimental procedures have been designed to block cancer spread in animal and some human systems. This paper will review some general aspects of cancer spread and then examine experimental approaches that block specific components of the cancer dissemination process that could become clinically useful.

Mechanisms of cancer spread

Normal tissues in the body possess stable, organized patterns of cells. Cellular proliferation is usually confined to a specific region of the tissue, and the cells are confined within the parent tissue. Malignant tumors (cancers) possess cells that can spread beyond the confines of the original tumor. Cancer spreads by two major means: 1) invasion, the spread of cells into adjoining tissue, and 2) metastasis, the spread of cells to distant sites, usually via the bloodstream, lymphatic system, or through body spaces. A detailed discussion of these processes is given in Ref. (1).

Invasion involves expansion of cancer cells into surrounding tissues as the result of continuous division. Active cell movement also appears to be involved in invasive behavior, and defective cellular adhesion appears to facilitate invasive spread. The growth and movement of cancer cells, easily observed in tissue culture, are not blocked by cell-cell contact, as appears to be the case for normal cells. Cancer cells release proteases, glycosidases, and other enzymes that help them digest away surrounding tissues and extracellular matrices so that expansion of the cells into surrounding areas can occur.

Metastasis can generally be divided into several specific steps:

1. Detachment of cells from the primary tumor
2. Invasion of these cells into lymph vessels or
Figure 2 Spread of a primary tumor to secondary sites. In this case the primary cancer is a melanoma on the forearm (P). Once these cells enter the circulation, they can be transported to and establish secondary cancers in organs such as the brain (b), the lungs (Lu), the liver (Li) or the ovaries (O). Drawing is not intended to be an accurate representation of organ or vessel locations.

Figure 3 Extravasation of blood-borne cancer cells. Cancer cells (drawn in black) adhere to capillary inner wall within a clot that has formed. The cells penetrate the blood vessel, entering surrounding tissue where they grow, forming secondary tumors.

blood vessels or body spaces and dissemination of the cells to distant areas
3. In the case of blood and lymph disseminated cancers, arrest of cells in the vessels of distant organs
4. Invasion of the cells through the vessel walls and into the tissue of secondary sites
5. Growth of secondary tumors at the secondary sites

A detailed discussion of these processes is given in Ref. (1) (See Figures 1 and 2.)

It is evident from this discussion that metastasis can be broken down into a several-step process. Each step, therefore, could become a point at which potential control of metastasis might be achievable. It is this topic, the experimental control of metastasis, that will be discussed next.

Experimental control of metastasis

Studies in which windows were constructed in the ears of rabbits so that filming the events that occur during cancer spread could be accomplished revealed some interesting information that led to potentially significant therapeutic applications. Cancer cells in the circulation were observed first to adhere firmly to the capillary endothelium (inner wall). These cells became trapped within a clot. The walls of the capillaries appeared damaged and white blood cells penetrated the damaged capillaries. Cancer cells began also to penetrate the damaged capillaries, enter the surrounding tissue and grow, obliterating the capillaries (Figure 3). New capillaries grew into the area of the dividing tumor mass leading to the development of large secondary cancers.

Wood and colleagues administered anticoagulants to the experimental animals and found a significant reduction in the numbers of secondary tumors that formed. These results, having potential clinical applications in humans, strongly suggest that the step in metastasis in which clots form around tumor cells trapped in capillaries may protect the cancer cells, enabling them to invade the capillary walls (Figure 3). The clots may also serve as a network on which white blood cells can migrate and help to damage the capillary endothelium. This work, therefore, presents an important example of the potential control of cancer spread by interfering with a specific component of the metastatic process.

Another important aspect of cancer development is the ingrowth of blood vessels into the growing tumor. Without such vascularization, tumors cannot grow to more than a few millimeters in diameter.
Wood and colleagues found that new capillaries grew into tumors at the rapid rate of up to 1 mm each day. If tumors were separated from surrounding tissue with a Millipore filter, capillary growth still occurred in the surrounding tissue, suggesting that tumor cells produce a substance that can diffuse through a filter and stimulate capillary growth. Such so-called tumor angiogenesis factors have been isolated and purified from tumors and placental tissue.

If capillary ingrowth into tumors could be prevented, the development of primary and secondary tumors would be severely restricted. Inhibitors of angiogenesis have been isolated from tissues such as cartilage and the vitreous humor of the eye. These inhibitors are positively charged proteins that have not as yet been fully characterized because of their limited availability. Recently, however, Folkman and Taylor of the Children's Hospital Medical Center in Boston characterized a specific commercially available inhibitor of capillary ingrowth. The substance is protamine, an arginine-rich, positively charged protein of about 4300 daltons molecular weight that can be isolated from sperm. Protamine inhibited capillary growth during embryonic development, in certain immune and inflammation reactions, and in the growth of solid tumors.

Taylor and Folkman also showed that a combination of heparin, an anticoagulant, and an angiogenesis promoter, isolated from tumor tissue, caused extensive angiogenesis, far greater than the angiogenesis promoter alone. Folkman's group noted that heparin is secreted by mast cells of connective tissue that assemble around the tumor before new capillary ingrowth is initiated.

Protamine binds heparin and blocks the growth of blood vessels into tumors. Although quite toxic, protamine, when administered to laboratory animals, resulted in a 77% to 97% inhibition of lung metastases in mice. It is likely that the antitumor properties of protamine result from its heparin binding activity because it is not toxic to tumor cells growing in culture.

Protamine has been used in the treatment of breast cancers in humans and animals and in treating some other cancers in England, with limited success. Although control of tumor angiogenesis has not yet been achieved to the point of major clinical usefulness in the treatment of cancer, these studies again clearly suggest that metastasis and tumor growth can be inhibited by specific interference with a key step in the metastatic process—capillary ingrowth into developing cancers.

Many malignant cells secrete high levels of enzymes that degrade proteins. These proteases probably play an important role in causing detachment...
of cells from primary tumors, invasion of these cells to surrounding tissues, and through blood vessel linings. Recent work by Nicolson and colleagues has also demonstrated that highly metastatic cells digest their way through blood vessel components as they extravasate from the blood vessels into the surrounding tissue (Figure 3). Proteolytic enzymes involved in this process were found to be released in membrane vesicles at the tumor cell surface.\(^1\) Experiments by Oppenheimer and colleagues have shown that it is possible to block the action of proteases, preventing some of the consequence of these enzymes in causing tumor cell spread. A specific protease inhibitor, Pepstatin A, blocked the disaggregation of tumor cell clusters and increased tumor cell adhesiveness.\(^6\) Here again we see that cancer cell spread, at least experimentally, can be potentially blocked by interfering with a specific mechanism implicated in causing invasion and metastasis.

The cell surface is obviously of central importance in the metastatic process. All contacts between tumor cells themselves and with other cells occur at the cell surface; thus, it is understandable that much recent attention has been given to attempts at control of metastatic disease by perturbation of the cancer cell surface. For example, Nicolson and colleagues used tunicamycin, a drug that blocks synthesis of sugar chains that are transported to the cell surface and appear to be involved in mediating cell-cell interaction.\(^1\)\(^5\)\(^6\) Treatment of metastatic cancer cells with this drug inhibited metastasis, while removal of the drug returned full metastatic potential to the cells within a day.\(^1\)

Nicolson and Poste explored the role of the cell surface in metastasis in other ways. They found that highly metastatic cells often release small membrane vesicles that can be purified and fused to cells that do not metastasize very easily. When these vesicles were fused to the low metastatic potential cell lines, these cells metastasized to a far greater extent.\(^1\) Thus, it appears that the cell surface contains specific molecules that control the arrest of cancer cells in secondary organs. Nicolson, McGuire, and colleagues found that a specific molecule, termed oncofetal antigen, appears to play a central role in targeting arrest of a specific line of mouse tumor cells to the secondary organ—the liver. When injected into mice, these cancer cells (a line of mouse lymphoma) form many secondary tumors in the liver. If, however, the tumor cells are coated with fragments of antibodies directed against the oncofetal antigen, present on the surfaces of the cells, liver tumors do not form and the mice do not die.\(^1\) In addition, antibody-treated cells do not bind to liver cell clusters in culture, while cells without anti-
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body do. These results show that metastasis can be experimentally controlled by blocking a specific cell surface antigen that appears to be involved in allowing cancer cells to adhere to liver tissue.

Chemotherapy has not cured many metastatic cancers because most of the chemicals used lack the precise specificity that is required to kill all of the cancer cells but not normal cells. It is likely that new drugs will be developed that possess better specificity. Drugs could be coupled with antibodies directed against specific cancer cell antigens such as the oncofetal antigen described in this report, and drugs might also be incorporated into membrane vesicles that can be specifically fused to cancer cell surfaces. This report serves to emphasize that cancer metastasis can be controlled at the experimental level by interfering with specific components of the processes of invasion and metastasis. A complete program to eradicate cancer will develop not only from control at the metastatic level but will include improvements in the early diagnosis and prevention of the disease and a better understanding of its causes. Current knowledge concerning cancer's causes and how it may be prevented has been reviewed previously elsewhere.¹,⁴,¹¹

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